REMARKS/ARGUMENTS

Claims 81, 85, 87, 91, 104-115 are active in this case.

Support for the amendments to Claims 81, 85, 87 and 91 is found on pages 7-8, page 17, pages 19-20, and the discussion of detecting two, three or more antigens by cocktailing the primary antibodies. Support for Claims 104-107 is on pages 9-11.

Support for Claims 108 and 109 is found in the paragraph bridging pages 7-8.

Support for Claims 110-115 is found in the specification on page 20, lines 2-4: "In a particularly preferred embodiment, the sample is treated with a device according to the description in U.S. patent no. 6,580,056, the entire contents of which are incorporated herein by reference." See also, e.g., MPEP § 2163.07(b).

No new matter is believed to have been added by the amendments.

Applicant thanks the Examiner for the courtesy of discussing this case with the undersigned on July 3, 2007. The substance of this discussion is summarized and expanded upon in the remarks below.

As discussed in the specification, double and triple stain technology using immunohistochemistry in formalin-fixed paraffin-embedded tissues has also been used for many years. Double stains are accomplished by applying primary antibodies and detection in a sequence of steps to achieve multiple labeling on the same tissue. Those methods apply the antibodies in series and are severely limited in terms of cost, time efficiency, as well as sensitivity vs. background signals.

As discussed on page 3 of the application, Illustrative of these disadvantages is the typical procedure of the previous technology to perform a double stain immunoassay. For example, the sample is treated with Hydrogen Peroxide for 5 minutes followed by two optional protein block (5 to 10 minutes) and Avidin-Biotin block (20 to 40 minutes). Then, the primary antibody is applied for 30 to 60 minutes, linked for 10 to 20 minutes, labeled

with, e.g., HRP for 10 to 20 minutes, treated with DAB for 5 minutes, and then denatured for 5 minutes. Subsequently, the second primary antibody is applied for 30 to 60 minutes, followed by an optional protein block for 5 to 10 minutes, linking for 10 to 20 minutes, and then labeling the second primary antibody, for example, with AP, for 10 to 20 minutes. The reactivity is detected by applying Fast Red for 10 to 20 minutes followed by counterstaining (plus bluing) for 30 and 60 seconds and coverslipping, which requires a water-base mounting media. The total time for this double stain procedure is about 3 to 4 hours with a total of 11-15 manual steps; plus 12 to 14 washes (32 maximum steps). Further, for a triple stain one would add 5 more steps (40+ maximum steps) and would take a total time of 4 to 5 hours.

The inventor, to the best of his knowledge, was the first to cocktail primary antibodies in a manner that the primary antibodies were stable, generated sensitive results (also with lower background than had ever been achieved before) with fewer steps and a time savings not only attributed to automating the staining process. In fact, automation of a simultaneous triple stain or more as is defined in the claims was not done nor could have been done because the reagents and methodologies as described and claimed in this application were not available.

Therefore, from the remarks below, it should be apparent that the cited prior art does not describe nor make obvious the claimed method because the prior art does not and, in fact, does not enable, the simultaneous detection of at least three antibodies using a single cocktail of at least three primary antibodies.

The rejection under 35 USC 103(a) based on van der Loos, the specification, Mers or Hasui is traversed.

While van der Loos generally discloses detecting two antigens simultaneously (starting at page 13) and immunoenzyme triple staining (Chapter 8, starting at page 63), he

does not describe simulatenously applying a cocktail of three primary antibodies to achieve that desired result. Indeed, as discussed by van der Loos on page 64, sec. 8.2 the third antibody is applied after the two antibodies have been applied so as to confirm the original double stain. Indeed, van der loos makes it quite clear in the following sec. 8.3 that triple stains have been rarely applied and published. This isn't surprising because they didn't work very well, gave poor results and were incredibly time consuming.

The Meyers paper does discuss automated staining using at least two antibodies but based on my understanding of page 109 and table 2 of Meyers the method used is a sequential application of the antibodies and not a simultaneous double-stain protocol, through a 137 step sequential protocol. Hasui is also relied upon to teach automated staining.

Again, what the prior art does not describe is the simultaneous detection of antigens using a cocktail that is applied to a sample and which contains three antibodies. Therefore, regardless of whether the art taught and/or the specification states that certain antigen combinations could be detected, the cited art does not describe or suggest the method as claimed. Furthermore, it is well-established law that in order for a reference to anticipate a claimed invention, the reference or references must provide an enabling disclosure sufficient to place the public in possession of the claimed invention.[1] Likewise, this analysis extends to obviousness, where a holding of obviousness cannot be sustained "unless there is some known or obvious way to make the thing or to carry out the process."[2]

The cited art does not provide an enabling disclosure that constitutes anticipation or obviousness to the claims of the present application because they give no guidance whatsoever as to how to successfully perform an at least triple stain on a given sample in an

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[1]See MPEP 2121.01 and *In re Hoeksema*, 399 F.2d 269, 158 USPQ 596 (CCPA 1968).

[2]See *In re Collins*, 462 F.2d 538, 174 USPQ 333 (CCPA 1972), citing *In re Hoeksema*, see *supra*.

automated staining device as claimed with the advantages and sensitivity that has been discussed above and in the present specification.

Accordingly, withdrawal of this rejection is requested.

The rejection under 35 USC 103(a) in view of Mason and Shi is also not applicable to the claims because while Mason does appear to describe a simultaneous double-staining protocol and the Shi et al reference describes a conjugate with poly HRP-linker, this cited art does not describe is the simultaneous detection of antigens using a cocktail that is applied to a sample and which contains three antibodies in an automated staining device as is claimed.

Accordingly, withdrawal of this rejection is requested.

The rejection under 35 USC 112, second paragraph is no longer applicable in light of the amendments submitted.

A Notice of Allowance for all pending claims is requested.

Respectfully submitted,

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